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(54) Title: COLLOIDAL NANOSILVER SOLUTION AND METHOD FOR MAKING THE SAME

(57) Abstract: The present invention provides a nanosilver composition which contains nanosilver particles having diameters between 1 nm and 100 nm. The silver content in the nanosilver composition is between 0.001% to 0.4% by weight. The nanosilver composition also contains a stabilizing agent which includes, but is not limited to, starch or its derivative, cellulose or its derivative, polymer or copolymer of acrylate or acrylate derivative, polyvinyl pyrrolidone, alginic acid, and xantham gum. The present invention also provides a method for making the nanosilver composition. The nanosilver composition prepared by this method does not contain any toxic substances.

COLLOIDAL NANOSILVER SOLUTION AND METHOD FOR MAKING THE SAME

FIELD OF THE INVENTION

The present invention relates to nanotechnology. In particular, the present invention relates to a method of producing silver particles in the nanometer range and the product produced therefrom.

DESCRIPTION OF THE RELATED ART

It has been known for centuries that silver possesses germicidal properties and has been employed as germicide before modern antibiotics were developed. During those days, users would shave silver particles into their drinking water, or submerge whole silver pieces in the drinking water, for the purpose of ingesting silver by drinking the water. Even after the onset of modern antibiotics era, silver remains to be used as antimicrobial agent, particularly because microorganisms treated by silver do not acquire resistance to the metals, while the conventional antibiotics often induce the formation of antibiotic-resistant microorganisms.

Silver is a safe and effective antimicrobial metal. In the late eighteenth century, western scientists confirmed that silver, which had been used in oriental medicine for centuries, was an effective antibacterial agent. Scientists also knew that the human body fluid is colloidal. Therefore, colloidal silver had been used for antibacterial purposes in the human body. By the early nineteenth century, colloidal silver was considered the best antibacterial agent until the discovery of the antibiotics. Due to the potency and revenue-driven advantages of antibiotics, the antibiotics gradually substituted colloidal silver as the main choice for antibacterial agent. However, thirty years into the discovery of the antibiotics, scientists began to discover that antibiotics induced the development of antibiotic-resistant bacterial strains which significantly affected the efficiency of antibiotics. Therefore, since 1870s, silver has again been recognized

as a preferred antibacterial use, particularly due to its non-toxic and non-induction of bacterial-resistant characteristics.

There are many reasons why administering silver suspended in solution (e.g., colloidal silver) would enhance an individual's health. It is possible that such a solution operates to inhibit the growth of bacteria, fungi, viruses, and other unwanted organisms, as well as eradicating such existing bacteria, fungi, viruses, and other organisms. It is also possible that a solution of silver can have an anti-inflammatory effect, sufficient to reduce symptoms of asthma. Silver in solution might also act in a similar fashion to a homeopathic remedy.

There have been numerous attempts to produce silver-based solutions, including colloidal silver. However, many of the silver-based products fail to maintain the silver particles in suspension, either because the silver solution is not a true colloid or because it is otherwise unstable. When the suspension of the silver particles fails, the particles fall to the bottom of the solution, thereby reducing the solution's concentration of silver and rendering it less effective.

For example, it is known that nanosilver particles in the one to one hundred nanometer diameter range may be produced by electrolysis. As explained above, such silver particles may be produced, but the stability is rather poor, with the particles increasing in size and precipitating out of solution within a few days.

It is therefore an object of the present invention to provide an improved method of producing silver particles in the nanometer range and compositions produced therefrom.

SUMMARY OF THE INVENTION

According to one aspect of the present invention, a nanosilver composition is provided containing nanosilver particles with diameter range of less than 100nm (e.g. ranged between one to one hundred nanometer) and remaining in that same diameter range for at least 110 days. That is to say, the 1 – 100 nm nanosilver particles do not react chemically with each other or merge physically with each other to become larger particles during at least 110 days. One of the examples illustrated below demonstrated that the resulting nanosilver particles are less than 35nm (e.g. sized between 1 to 35 nm) with at least 30% of the nanosilver particles with diameters of less than 15nm (e.g. 1 to 15 nm) for at least 110 days.

In the most preferred embodiment, the composition according to the present invention further comprises at least 0.1% weight by weight of a stabilizing agent. This stabilizing agent has the ability to slightly increase the viscosity of the reaction solution that is used to produce the composition according to the present invention. Due to this slight increase in viscosity, the nanosilver particles produced therefrom do not react with each other again to form larger particles and therefore remain stable for at least 110 days.

As a working example, the stabilizing agent may be used in the range of 0.1 to 5%, preferably 0.2 to 3% weight by weight.

In another aspect of the present invention, a nanosilver gel composition is provided with nanosilver particles in the range of less than 100nm (e.g. one to one hundred nanometers) and further comprising a gelling agent to increase the viscosity of the composition according to the users' requirement. Such a composition is again stable for at least 110 days and preferably also contains a stabilizing agent.

In one embodiment, the stabilizing agent may be starch or its derivative, cellulose derivatives, polymer or copolymer of acrylate or acrylate derivative, polyvinyl pyrrolidone, alginic acid, or xanthan gum. Examples of starch derivative include, but are not limited to, sodium carboxymethyl starch, hydroxyethyl starch, and pregelatinized starch. Examples of cellulose derivatives include, but are not limited to, methyl cellulose, hydroxypropyl cellulose, hydroxypropyl methyl cellulose, and hydroxyethyl cellulose. An example of polymer or copolymer of acrylate derivative is carbomer, which is a carboxy vinyl polymer. Carbomer generally are high molecular weight ("MW") polymers (MW above 1,000,000). Carbomer is commercially available. B. F. Goodrich Company currently sells carbomer using the tradename of Carbopol. Carbopol 934P has a MW of about 3,000,000 and Carbopol 940 is about 4,000,000. The preferred concentration of the stabilizing agent is at about 0.2 to 5% by weight of the total solution. The more preferred range is 0.2 to 3%.

The nanosilver composition has antimicrobial activity, particularly for inhibiting growth of bacteria, fungi, or chlamydia. Examples of microorganisms which can be inhibited by the colloidal nanosilver solution include, but are not limited to, *Escherichia coli, Methicillin resistant Staphylococcus aureus, Chlamydia trachomatis, Providencia stuartii, Vibrio vulnificus, Pneumobacillus, Nitrate-negative bacillus, Staphylococcus aureus, Candida albicans, Bacillus cloacae, Bacillus allantoides, Morgan's bacillus (Salmonella morgani), Pseudomonas maltophila, Pseudomonas aeruginosa, Neisseria gonorrhoeae, Bacillus subtilis, Bacillus foecalis alkaligenes, Streptococcus hemolyticus B, Citrobacter,* and *Salmonella paratyphi C.*

According to another aspect of the present invention, there is a provided method for making a nanosilver composition. The method includes the following steps: (1) dissolving Silver Oxide (Ag_2O) in ammonia water ($NH_3 \cdot H_2O$) to form a silver ammonia complex ion [$Ag(NH_3)$

$_2^+$] -containing solution; (2) adding a stabilizing agent to the silver ammonia complex ion-containing solution to increase the viscosity and to form a stabilized silver ammonia complex ion solution; and (3) adding a reducing agent to the stabilized silver ammonia complex ion-containing solution to form nanosilver particles of 1-100nm diameter. Without being bound to any theory, the inventors believe that the stabilizing agent functions as a means to slightly increase the viscosity of the silver ammonia complex ion-containing solution such that nanosilver particles of less than 100nm (e.g. in the range of one to one hundred nanaometer) formed therefrom would remain stable and the diameter staying below 100nm for at least 110 days.

The stabilizing agent may be any polymer as described above or any other compound that increases the viscosity of the silver ammonia complex ion-containing solution slightly in order to prevent the destabilization of the silver crystals after formation. Numerous examples will be described in the following section to teach one in the art to produce this composition. Furthermore, the use of the various polymers or other compounds as a stabilizing agent and the suitable concentrations therefor are disclosed. In addition, Examples 7 and 8 teach how to examine the products for the size or diameter of the nanosilver particles produced and how to test for antimicrobial activity respectively. Therefore the use of other compounds as stabilizing agent and in the appropriate concentrations that are not described in the following examples are not precluded in the claims, as they may be determined by one skilled in the art without undue experimentation based on the teaching provided herein. Furthermore, other methods for testing the final products after the reaction to determine the size of the nanosilver particles are within the knowledge of one skilled in the art. To test for the stability of the particles after formation over time, the same test may be performed over time.

Many compounds may be used as the reducing agent in the instant invention including but not limited to glucose, ascorbic acid, hydrazine hydrate and hydrogen peroxide. For a pharmaceutical composition, hydrazine hydrate is one preferred reducing agent because it may react with air to form harmless substances that would not affect the human body.

Also in the most preferred embodiment, silver oxide is used to produce the silver ammonia complex ion as the silver particle produced by the reduction of silver oxide according to the present invention produces more stable nanoparticles than those reduced by other salt of silver such as silver nitrate.

By way of example, the ammonia water used in step (1) is preferred to be at a concentration of 25% to 28% by weight and more preferably at concentration of 28%. Also, the silver oxide and the ammonia water in step (1) is preferred to be at a ratio of about 1:5 to about 1:10 (w/w). In addition, the silver oxide and the hydrazine hydrate in step (4) is preferred to be at a ratio of about 1:0.087 to about 1:0.26 (w/w).

In one embodiment, the stabilized silver ammonia complex ion-containing solution is mixed with hydrazine hydrate ($NH_2NH_2 \cdot H_2O$) at about 0 to 45°C for about 0.5 to 2 hours. Also, after the formation of the nanosilver composition, it is preferred to let the colloidal nanosilver solution be in contact with air for about 0.5 to 5 hours.

The nanosilver composition can be used as an antibacterial or antifungal agent for treatment of patients with burn and scald-related skin infection, wound-related skin infection, dermal or mucosal bacterial or fungal infection, surgery cut infection, vaginitis, and acne-related infection, by applying or spraying the solution onto the wounds. It can also be used as a disinfectant or sanitary agent to clean areas in need of disinfection or sanitation.

The nanosilver composition of the present invention can maintain the colloidal nanosilver particles in suspension for a long period of time and it is particularly suitable for industrial purpose. It also has the advantages of not containing toxic substances so that it is particularly suitable for medicinal and healthcare use.

DETAILED DESCRIPTION OF THE INVENTION

The following is the description of the preferred embodiments of the present invention. The silver nanoparticles produced according to the instant method and in the product according to the present invention are generally described as being in the range of one to hundred nanometer. It is understood that this range is used for ease of description, and that silver particles of less than 1nm would also have antimicrobial activity and is not precluded from such a description.

In the preferred embodiment, the nanosilver composition of the present invention is further characterized by being non-toxic and its stability. The silver content in the instant nanosilver composition may be between 0.001% to 0.4% by weight. It is also stable at room temperature (about 25°C or 77°F) for at least 110 days. Because of these characteristics, the nanosilver composition is particularly suitable for use in healthcare related matters such as sanitization, disinfection and for industrial purposes.

The nanosilver composition of the present invention can be used in sanitary products, which include, but are not limited to, solutions for cleansing agents for clothing, women hygiene, acne or pimples, and soaking solution for tooth brush. It can also be used in healthcare products, which include, but are not limited to, treating patients with all kinds of injuries and/or burns, bacterial and fungal infections (including gynecological infections such as vaginitis), gastrointestinal bacterial infection, and sexually transmitted diseases. In addition, the nanosilver composition of the present invention can be used in industrial products, which include, but are not limited to, food preservatives especially for fruits and vegetables, drinking water disinfectants, paper and construction filling materials preservation (especially to prevent mold formation).

The nanosilver composition of the present invention possesses a broad spectrum of antibacterial and antifungal ability. It can kill and suppress growth of bacteria and fungi, such as *Escherichia coli, Methicillin resistant Staphylococcus aureus, Chlamydia trachomatis, Providencia stuartii, Vibrio vulnificus, Pneumobacillus, Nitrate-negative bacillus, Staphylococcus aureus, Candida albicans, Bacillus cloacae, Bacillus allantoides, Morgan's bacillus (Salmonella morgani), Pseudomonas maltophila, Pseudomonas aeruginosa, Neisseria gonorrhoeae, Bacillus subtilis, Bacillus foecalis alkaligenes, Streptococcus hemolyticus B, Citrobacter,* and *Salmonella paratyphi C.*

The antibacterial and antifungal activity of the nanosilver composition of the present invention has advantage over the conventional antibiotics in killing and suppressing bacterial growth, as it does not induce drug-related resistance in the bacterial or fungal strains.

Using conventional preparation methods, other compounds besides silver are generated such as, oxidized products of the reducing agents, which are possibly toxic. The presence of these oxidized products not only affect the purity of the product but also make it unsuitable for use in healthcare related industry due to its toxicity. Also, although the oxidized products of the reducing agents can be removed subsequently by conventional methods, such as dialysis, the method of dialysis involves excessive steps which not only is time-consuming but also adds more difficulties and expenses to the industrial-scale manufacturing process.

To avoid producing unwanted toxic products, at least two methods are disclosed in the prior art which produce a silver containing solution without harmful side products from the reducing agents. For example:

(1) Reacting silver oxide (Ag_2O) with hydrogen gas to form metallic silver and water:

$Ag_2O + H_2$? $2\ Ag + H_2O$

(2) Reacting silver oxide (Ag_2O) with hydrazine hydrate ($NH_2NH_2 \cdot H_2O$) to form metallic silver, nitrogen gas, and water.

$2\ Ag_2O + NH_2NH_2 \cdot H_2O$? $4\ Ag + N_2 + 3\ H_2O$

Because the above reactions produce metallic silver, nitrogen gas, and water, which are non-toxic in nature so that no additional steps are necessary for removing the unwanted toxic products, theoretically, they should be suitable for the production of colloidal silver solution. However, the reactions as shown above are not practical in manufacturing industrial-scale l nanosilver composition. For example, in the reaction as described in (1), which requires the silver oxide to interact with hydrogen gas, a multiphase reaction is involved which makes it very difficult to carry out. See V. Kohlschuetter Strassburg (Z. Elektrochem., 14, 49-63. CA: 2: 1379-1380). When the silver oxide and hydrogen are sealed in a glass tube and reacted at 18°C or lower, the reduction reaction takes place very slowly. On the other hand, if the reaction is carried out at 60 °C or lower, the hydrogen gas is discharged into the saturated silver oxide solution, which results in yielding a colloidal silver solution with silver particles partially in suspension and partially precipitated. A colloidal silver solution prepared in this way is not suitable for use in sanitation or healthcare products due to precipitation of silver.

Also, in the reaction as described in (2) above, the interaction of silver oxide with hydrazine hydrate in water is limited by the low solubility of the silver oxide in water. See J. Voigt et al. (Z. Anorg. Allgem. Chem. 164, 409-419, CA21:3512). Therefore, in order to obtain a soluble silver oxide solution, the silver content of the silver oxide solution must be no more than 0.001% by weight. Using such a diluted silver oxide solution as starting material, the

resulting silver content in the colloidal silver solution is too low to be effective for sanitation or healthcare use.

The present invention provides a method for making a nanosilver composition which is distinctively different from the above-mentioned prior art methods. Based on this method, a nanosilver composition which contains high silver concentration (i.e., containing 0.001% to 0.4% by weight of silver), high stability (i.e., stable at room temperature for no less than 110 days), and no toxic substances, is formed.

The method for preparing the nanosilver composition of the present invention contains the following reactive steps:

(1) Dissolution of Silver Oxide in Ammonia Water ($NH_3 \cdot H_2O$).

Silver oxide (Ag_2O) is dissolved in ammonia water ($NH_3 \cdot H_2O$) to obtain a silver ammonia complex ion $[Ag(NH_3)_2^+]$ solution where the silver ion is in the form of silver ammonia complex ion$[Ag(NH_3)_2^+]$ as follows:

$$Ag_2O + 4\ NH_3 \cdot H_2O \rightarrow [Ag(NH_3)_2^+] + 2OH^- + 3\ H_2O$$

The concentrated ammonia water is preferred to be about 25 to 28% and most preferred concentration is at 28%. The preferred ratio of silver oxide and concentrated ammonia water is at about 1:5 to about 1:10, w/v. This procedure has the advantage of increasing the solubility and concentration of silver in the solution.

(2) Dissolution of stabilizing Agent in Water to Form a Viscous Medium.

A viscous medium is provided by dissolving a stabilizing agent in water. This viscous medium serves as a protective colloid mechanism for keeping the nanosilver particles with diameter in 1 – 100 nm range in the nanosilver composition and preventing the nanosilver

particles from aggregating with each other to form bigger particles and precipitating out of the solution. In the 1 – 100nm range the silver particles remain suspended in a colloidal state, and have very high biocidal activity. Preferably, the concentration of the stabilizing agent is between 0.2% to 5% by weight.

The stabilizing agent can be a synthetic or natural polymer or a combination thereof, which can be readily dissolved in water. Examples of the stabilizing agent include, but are not limited to, starch or starch derivatives, cellulose derivatives, polymer or copolymer of acrylate or acrylate derivatives, polyvinyl pyrrolidone (PVP), alginic acid, and xanthan gum. The starch derivatives include, but are not limited to, sodium carboxymethyl starch, hydroxyethyl starch, and pre-gelatinized starch. The cellulose derivatives include, but are not limited to, methyl cellulose, hydroxypropyl cellulose, hydroxypropyl methyl cellulose, and hydroxyethyl cellulose. The polymer or copolymer of acrylate or acrylate derivative is preferred to be Carbomer.

Carbomer is a polymer of acrylic acid (or a carboxy vinyl polymer). It is currently sold under the tradename of Carbopol by B.F. Goodrich Company. The preferred carboxy vinyl polymer is a high molecular weight (preferably MW above 1,000,000; and most favorably MW above 3,000,000) polymer, such as Carbopol 934P which has a molecular weight of about 3,000,000.

Carbopol is the trademark of B. F. Goodrich Company's carboxy vinyl polymers, which generally are high molecular weight ("MW") polymers (MW above 1,000,000). Specifically, Carbopol 934P has a MW of about 3,000,000 and Carbopol 940 is about 4,000,000.

(3) Mixing Silver Ammonia Complex Ion Solution with the Viscous Medium.

The silver ammonia complex ion $[Ag(NH_3)_2^+]$ solution is thoroughly mixed with the viscous medium to form a uniformly dispersed stabilized silver ammonia complex ion solution

to be used for the next reaction. The stabilized silver ammonia complex ion solution is preferred to be controlled at about 0° to 45°C.

(4) Reaction of Stabilized Silver Ammonia Complex Ion with Hydrazine Hydrate.

The stabilized silver ammonia complex ion is further interacted with hydrazine hydrate in the presence of oxygen gas to form metallic silver, nitrogen gas, and water as follows:

$$4Ag(NH_3)_2^+ + 4OH^- + NH_2NH_2H_2O + 6\,H_2O \;?\; 4\,Ag + 8NH_3H_2O + N_2$$

The preferred temperature for the above reaction is at about 0°C-45°C. The reaction is preferred to be conducted in about 0.5 to 2 hours. The silver oxide and hydrazine hydrate are preferred to be at a ratio of 1:0.087 to 1:0.26 by weight. The nanosilver particles prepared by the reactive steps (1)-(4) have diameter of 1 nm to 100 nm.

Because hydrazine hydrate is toxic, after the completion of step (4), the nanosilver composition is preferred to be kept in the presence of air for additional 0.5 to 5 hours so that the residue of hydrazine hydrate in the nanosilver composition containing solution can be decomposed into nitrogen and water by the following oxidative reaction:

$$NH_2NH_2{\cdot}H_2O + O_2 ?\; N_2 + 3\,H_2O$$

The resulting nitrogen gas and water are non-toxic so that no removal of the side products is necessary.

Moreover, the present invention provides a method for making the nanosilver composition of high silver concentration, high stability. If hydrazine hydrate is used as the reducing agent, no toxic ingredients will be produced. The above mentioned problems associated with the reaction are solved in the present invention: the solubility of silver oxide and final concentration of silver in the colloidal solution are improved, the nanosilver is stabilized in a colloidal state, and the toxic reactant, hydrazine, is carefully removed from the nanosilver

composition containing solution by further decomposition reaction with oxygen in the air. The nanosilver composition of the present invention is suitable for healthcare purposes and serves as an effective antimicrobial agent.

The "nanosilver composition" as described in the present invention is also referred to as a colloidal nanosilver solution in the following examples.

EXAMPLE 1

Preparation of the Colloidal nanosilver Solution of the Present Invention

The colloidal solution containing nanosilver particles of the present invention was prepared according to the following steps:

1. 60 g of sodium carboxymethyl starch was dissolved in 1600 ml of distilled water. The dissolved sodium carboxymethyl starch solution was added to a 5000 ml flask. The flask was heated to and maintained at 70°C until the solution became gelatinized. The resultant viscous medium was cooled down to room temperature.

2. 3 g of silver oxide was added to and mixed with 22 g of 28% ammonia water in 115 ml of distilled water to form a silver ammonia complex ion (i.e., $[Ag(NH_3)_2^+]$) -solution.

3. The silver ammonia complex ion solution was then mixed thoroughly with the viscous medium of (2) to form a stabilized silver ammonia complex ion solution.

4. 0.5 g of 80% hydrazine hydrate was mixed with and dissolved in 200 ml of distilled water to form a hydrazine hydrate solution.

5. The hydrazine hydrate solution was, with stirring, added to the flask containing the stabilized silver ammonia complex ion solution. The solution was then reacted at room temperature for 4.5 hours.

EXAMPLE 2

Preparation of the Colloidal Nanosilver Solution of the Present Invention

The colloidal solution containing nanosilver particles of the present invention was prepared according to the following steps.

1. 1600 ml of distilled water was added to a 5000 ml flask and heated to and maintained at 70°C.

2. 50 g of methyl cellulose was gradually added to the flask containing the heated distilled water. After thorough mixing of the methyl cellulose with the distilled water, the temperature of the solution was gradually reduced to around 30 °C until a viscous medium was formed.

3. 3 g of silver oxide was added to and mixed with 22 g of 28% ammonia water in 125 ml of distilled water to form a silver ammonia complex ion solution.

4. The silver ammonia complex ion solution was then added to and mixed with the viscous medium to form a stabilized silver ammonia complex ion -solution.

5. 0.6 g of 80% hydrazine hydrate was dissolved in 200 ml of distilled water to form a hydrazine hydrate solution.

6. The hydrazine hydrate solution was then added to the flask containing stabilized silver ammonia complex ion solution. The solution was then reacted at room temperature for 5 hour under seal.

EXAMPLE 3

Preparation of the Colloidal Nanosilver Solution of the Present Invention

The colloidal solution containing nanosilver particles of the present invention was

prepared according to the following steps.

1. 1600 ml of distilled water was added to a 5000 ml flask.

2. 10 g of polyvinylpyrrolidone (PVP) was gradually added into the flask at room temperature and dissolved therein to form a viscous medium.

3. 6 g of silver oxide was dissolved in 44 g of 28% ammonia water in 70 ml of distilled water to form a silver ammonia complex ion solution.

4. The silver ammonia complex ion solution of step (3) was add to and thoroughly mixed with the viscous medium of (2) to form a stabilized silver ammonia complex ion - solution.

5. 1.5 g of 80% hydrazine hydrate was dissolved in 270 ml of distilled water to form a hydrazine hydrate solution.

6. The hydrazine hydrate solution was then mixed with the stabilized silver ammonia complex ion -solution of (4) in the flask. The flask was kept at 30 °C and stirred for 5 hours.

EXAMPLE 4

Preparation of the Colloidal Nanosilver Solution of the Present Invention

The colloidal solution containing nanosilver solution of the present invention

The colloidal solution containing nanosilver particles of the present invention was prepared according to the following steps.

1. 1755ml of distilled water was added to a 5000ml flask and heated to and maintained at 70°C.

2. 36g of hydroxypropyl methyl cellulose was gradually added to the flask containing the heated distilled water. After thorough mixing of the hydroxypropyl methyl cellulose with the distilled water, the temperature of the solution was gradually reduced to around 30°C until a viscous medium was formed.

3. 4.5g of silver oxide powder was added to and mixed with the solution of 25g of 25% ammonia water in 80ml of distilled water to form a silver ammonia complex ion solution .

4. The silver ammonia complex ion solution was then added to and mixed with the viscous medium to form a stabilized silver ammonia complex ion-viscous medium.

5. 0.5g of 80% hydrazine hydrate was dissolved in 100ml of distilled water to form a hydrazine hydrate solution.

6. The hydrazine hydrate solution was, with stirring, added to the stabilized silver ammonia complex ion –solution .

7. The stirring was maintained for 4 hours to allow the solution to be in touch with air to obtain the colloidal nanosilver solution of the present invention .

EXAMPLE 5

Preparation of the Colloidal Nanosilver Solution of the Present Invention

The colloidal solution containing nanosilver solution of the present invention

The colloidal solution containing nanosilver particles of the present invention was prepared according to the following steps.

1. 927 ml of distilled water was added to a 2000ml flask and heated to and maintained at 70°C.

2. 20 g of hydroxypropyl methyl cellulose was gradually, with stirring added to the flask containing the heated distilled water after through mixing of the hydroxyproyl methyl cellulose with the distilled water, the temperature of the mixture was gradually reduced to around 30 °C until a viscous medium is formed

3. 2.2 g of silver oxide powser was added to and mixed with the solution of 13g of 25% ammonia water in 25 g of distilled water to form a silver ammonia complex ion solution.

4. The silver ammonia complex ion solution was then added to and mixed with the viscous medium to form a stabilized silver ammonia complex ion solution.

5. 1.3g of 30% hydrogen peroxide was dissolved in 13g of distilled water.

6. The hydrogen peroxide solution was with stirring, added to the stabilized silver ammonia complex ion solution at room temperature. The stirring was maintained for 1 hour to obtain the colloidal nanosilver solution of the present invention.

EXAMPLE 6

Preparation of the Nanosilver of the Present Invention in Gel State

The colloidal solution containing nanosilver particles of the present invention was prepared according to the following steps.

1. 10g of carbomer was added gradually to 700ml of distilled water and maintained in room temperature for 24 hours after adding.

2. 30g of Glycerol was added and stirred thoroughly.

3. 143g of colloidal nanosilver solution from example 4 was added and continued to stir for another 0.5 hour

4. 5g of 28% ammonia water in 100ml distilled water was added gradually with stirring, the pH of the mixture was adjusted to pH6.5 to pH7. Distilled water was then added to the mixture until the total mass reached 1000g and continued to stir for another 1 hour. Distilled water was then added to the mixture until the total mass reached 1000g and continued to stir for another 1 hour. The nanosilver composition in gel state is obtained.

EXAMPLE 7

Examination of the Dimension and Stability of the Colloidal nanosilver Solution

I. Purpose:

The colloidal solution containing nanosilver particles of the present invention was examined for the dimension of the nanosilver particles and stability of the nanosilver composition over time (days) in terms of suspension by electron microscopy.

II. Method:

In accordance with the standard procedures for JY/T011-1996 transmission electron microscope, JEM-100CXII transmission electron microscope was used under the testing conditions of accelerating voltage at 80 KV and resolution at 0.34 nm. The colloidal nanosilver solutions produced by Examples 1-4 of the present invention were observed for the size and distribution of the nanosilver particles therein. Aliquots of the samples from Examples 1-4 were taken out from the solutions either being freshly made or after being stored at room temperature for 110 days.

III. Results:

For the freshly made nanosilver samples, the diameters of all the silver particles contained therein were below 35 nm, among which, most particles (37%) had a diameter of 15 nm.

For the colloidal solution stored after 110 days, the diameters of all the silver particles contained therein were kept below 35 nm, among which, most particles (38%) had a diameter of 15 nm.

IV. Conclusion:

The colloidal solution of the present invention containing nanosilver particles which had a size range of 1 nm to 100 nm and was very stable after storage of 110 days at room temperature. There was no visible increase in size of the silver particles contained therein and no precipitation of silver particles. The colloidal solution of the present invention was stable for further processing and adopted for use, storage, and transportation.

EXAMPLE 8

Antimicrobial Activity of the Colloidal Nanosilver Solution of the Present Invention

I. Purpose:

The colloidal solution of the present invention was tested for the antimicrobial ability.

II. Method:

Microbial strains tested were *Escherichia coli, Methicillin resistant Staphylococcus aureus, Chlamydia trachomatis, Providencia stuartii, Vibrio vulnificus, Pneumobacillus, Nitrate-negative bacillus, Staphylococcus aureus, Candida albicans (ATCC 10231), Bacillus cloacae, Bacillus allantoides, Morgan's bacillus (Salmonella morgani), Pseudomonas maltophila, Pseudomonas aeruginosa, Neisseria gonorrhoeae, Bacillus subtilis, Bacillus foecalis alkaligenes, Streptococcus hemolyticus B, Citrobacter,* and *Salmonella paratyphi C.* These strains were either isolated from clinical cases or purchased as standard strains from Chinese Biological Products Testing and Standardizing Institute.

A typical example of the test, as illustrated by *Candida albicans* (ATCC 10231), was as follows:

Colloidal nanosilver solutions of examples 1-4 (each contains a concentration of 1370 μg/ml of silver) were tested for its antifungal activity against *Candida albicans*. The colloidal nanosilver solutions were diluted in distilled water to make the final concentrations of 137 μg/ml, 68.5 μg/ml, 45.7 μg/ml, 34.2 μg/ml, and 27.4 μg/ml. In the control group, no colloidal nanosilver solution was added. *Candida albicans* was added to each tested and control groups, respectively, and the viability of the fungus in each group was examined 2 minutes after incubation with the colloidal nanosilver solutions of examples 1-4.

Typically, due to the resilience of *Candida Albicans*, a higher concentration of disinfectant is required to kill or suppress the growth of *Candida albicans* than for killing bacteria such as *Staphylococcus aureus* and *Escherichia coli*.

III. Results:

There was an average of 99.99% killing rate (1.78×10^6 cfu/mu) for all of the colloidal nanosilver solution tested (Examples 1-4) after 2 minutes of incubation. Among the same example, the most diluted sample demonstrated about the same fungicidal activity as the least diluted one.

IV. Conclusion:

The colloidal solution containing nanosilver particles of the present invention was effective as antimicrobial agent even at a diluted concentration of 27.4 μg/ml of silver.

EXAMPLE 9

Preparation of Nanosilver composition for Topical usage

1. Mix nanosilver composition (prepared by any methods stated in Example 1 to Example 6) with emulsifying or gelling agent used in medical application.

2. The preparation can then be applied to wounds, inflamed area or ulceration for antimicrobial purpose.

EXAMPLE 10

Preparation of Nanosilver composition as antimocrobial coating agent

1. Mix nanosilver composition (prepared by any methods stated in Example 1 to Example 6) with spraying device or a glue to coat onto a desired surface..

2 The preparation can then be applied to coat any surface including fabric hard object and even smooth surfaces such as man- made yarn for antimicrobial purpose.

It is clear from the above non-limiting examples that numerous embodiments and variations may be produced and used according to the teaching provided herein. It is to be understood that the invention is not limited to the disclosed embodiments. On the contrary it is intended to cover modifications as would be apparent to those skilled in the art. Therefore, the scope of the appended claims should be accorded the broadest interpretation so as to encompass all such modifications. In particular, the stable nanosilver composition of the instant invention may be applied to any surfaces or materials either alone or in combination with other substances such as glues, gels or binders. The composition may also be used to produce sterilizing sprays, or coated onto various types of fabric and solid material in the presence or absence of added moisture. It may be used for the medical industry, construction industry or any other industry that benefits from nanosilver particles whether they be used for medical purposes or other uses not described herein. It is intended that the scope of the invention be defined according to the claims and their equivalents herein.

We claim:

1. A nanosilver composition comprising nanosilver particles with diameter less than 100 nm and remaining below 100nm diameter for at least 110 days at room temperature.

2. A nanosilver composition according to claim 1 wherein said further comprises all nanosilver particles have diameters of less than 50nm for at least 110 days.

3. A nanosilver composition according to claim 1 wherein at least 30% of silver particles have a diameter of less than 15nm for at least 110 days.

4. A nanosilver composition according to claim 1 further comprising at least 0.1%w/w of a stabilizing agent

5. A nanosilver composition according to claim 4 wherein said stabilizing agent is found in the range of 0.1 to 3% w/w.

6. A nanosilver composition according to claim 1 comprising 0.001% to 0.4% by weight of silver.

7. The nanosilver composition according to claim 1 further comprising a gelling agent to form a gel.

8. A nanosilver composition according to claim 7 wherein said gelling agent is present in the range of 0.2 to 5% w/w

9. A nanosilver composition according to claim 4 or 5 wherein said stabilizing agent is at least one polymer selected from a group consisting of starch, derivative of starch, derivative of cellulose, polymer of acrylate, copolymer of acrylate, acrylate derivative, polyvinyl pyrrolidone, alginic acid, andxanthan gum.

10. A nanosilver composition according to claim 9 wherein said derivatived starch is sodium carboxymethyl starch, hydroxyethyl starch, pregelatinized starch or dextrin; said derivatived cellulose is methyl cellulose, hydroxypropyl cellulose, hydroxypropyl methyl cellulose or hydroxyethyl cellulose; and said polymer/copolymer of acrylate/acrylate derivative is Carbopol.

11. A nanosilver composition according to claim 7 wherein said gelling agent is at least one polymer selected from a group consisting of starch, derivative of starch, derivative of cellulose, polymer of acrylate, copolymer of acrylate, acrylate derivative, polyvinyl pyrrolidone, alginic acid, and xanthan gum.

12. A nanosilver composition according to claim 11 wherein said derivatived cellulose is methyl cellulose, hydroxypropyl cellulose, hydroxypropyl methyl cellulose or hydroxyethyl cellulose; and said polymer/copolymer of acrylate/acrylate derivative is Carbopol.

13. A method for making a nanosilver composition comprising:

(a) dissolving silver oxide (Ag_2O) in ammonia water ($NH_3 \cdot H_2O$) to form a solution containing silver ammonia complex ion $[Ag(NH_3)_2^+]$;

(b) adding a stabilizing agent to said silver ammonia complex ion-containing solution to increase the viscosity and to form a stabilized silver ammonia complex ion solution; and

(c) adding a reducing agent to said stabilized silver ammonia complexion-containing solution to form nanosilver particles of 1-100nm diameter

wherein said viscosity in step (b) is sufficiently high to allow nanosilver particles of 1-100nm diameter to be formed in step (c) and remain in the 1-100nm diameter range for at least 110 days.

14. The method according to claim 13, wherein said stabilizing or gelling agent is at least one polymer selected from the group consisting of starch or starch derivative, cellulose derivative, polymer or copolymer of acrylate or acrylate derivative, polyvinyl pyrrolidone, alginic acid, and xanthan gum.

15. The method according to claim 14, wherein said cellulose derivative is methyl cellulose, hydroxypropyl cellulose, hydroxypropyl methyl cellulose, or hydroxyethyl cellulose.

16. The method according to claim 13, wherein said reducing agent is hydrazine hydrate or hydrogen peroxide.

17. The method according to claim 13 wherein said stabilized silver ammonia complex ion-containing solution is mixed with hydrazine hydrate ($NH_2NH_2 \cdot H_2O$) at about 0 to 45°C for about 0.5 to 5 hours.

18. The method according to claim 13 wherein said hydrazine hydrate is at 80% of concentration before mixing.

19. The method according to claim 13, wherein said ammonia water at ($NH_3 \cdot H_2O$) is at concentration of about 25% to 28% by weight.

20. The method according to claim 13, wherein said the concentration of ammonia water is at 28% by weight.

21. The method according to claim 16 further comprising a step of contacting said composition with air for about 0.5 to 5 hours after step (c).

22. The method according to claim 13, wherein said silver oxide and said ammonia water is at a ratio of about 1:5 to about 1:10, w/w.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SG 03/00062-0

CLASSIFICATION OF SUBJECT MATTER

IPC[7]: B01J 13/00, A10N 25/04, A01N 59/16, A61K 33/38

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC[7]: A01N, A61K, B01J

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

EPODOC, WPI, PAJ

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
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A	US 4181786 A (MUNE et al.) 1 January 1980 (01.01.80) *examples 1-7.*	1-6
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☒ Further documents are listed in the continuation of Box C. ☒ See patent family annex.

* Special categories of cited documents:
„A" document defining the general state of the art which is not considered to be of particular relevance
„E" earlier application or patent but published on or after the international filing date
„L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
„O" document referring to an oral disclosure, use, exhibition or other means
„P" document published prior to the international filing date but later than the priority date claimed
„T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
„X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
„Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
„&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
4 July 2003 (04.07.2003)	12 August 2003 (12.08.2003)
Name and mailing adress of the ISA/AT Austrian Patent Office Dresdner Straße 87, A-1200 Vienna Facsimile No. 1/53424/535	Authorized officer PUSTERER F. Telephone No. 1/53424/311

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SG 03/00062-0

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

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A	US 2002/0022012 A1 (COOPER et al.) 21 February 2002 (21.02.02) *claims 1,14&15; examples.*	1-6
A	US 5785972 A (TYLER) 28 July 1998 (28.07.98) *claims 1-3.*	1-6

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Information on patent family members

Interna[redacted]l application No.
PCT/SG 03/00062-0

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